

FILE 'HOME' ENTERED AT 09:14:57 ON 09 MAR 2009

```
=> index biosceince
'BIOSCEINCE' IS NOT A VALID FILE NAME
ENTER A FILE NAME OR (IGNORE):end
```

```
=> index bioscience
FILE 'DRUGMONOG' ACCESS NOT AUTHORIZED
COST IN U.S. DOLLARS
```

	SINCE FILE	TOTAL
	ENTRY	SESSION
FULL ESTIMATED COST	0.22	0.22

INDEX 'ADISCTI, ADISINSIGHT, ADISNEWS, AGRICOLA, ANABSTR, ANTE, AQUALINE, AQUASCI, BIOENG, BIOSIS, BIOTECHABS, BIOTECHDS, BIOTECHNO, CABA, CAPLUS, CEABA-VTB, CIN, CONFSCI, CROPB, CROPU, DDFB, DDFU, DGENE, DISSABS, DRUGB, DRUGMONOG2, DRUGU, EMBAL, EMBASE, ...' ENTERED AT 09:15:30 ON 09 MAR 2009

68 FILES IN THE FILE LIST IN STNINDEX

Enter SET DETAIL ON to see search term postings or to view search error messages that display as 0* with SET DETAIL OFF.

```
=> s ferrocen? (s) (prote6 or peptidase or hydrol4 or cleav3 or digest4)
UNMATCHED RIGHT PARENTHESIS '4)'
The number of right parentheses in a query must be equal to the
number of left parentheses.
```

```
=> s ferrocen? (s) (prote6 or peptidase or hydrol4 or cleav3 or digest4)
UNMATCHED RIGHT PARENTHESIS '4)'
The number of right parentheses in a query must be equal to the
number of left parentheses.
```

```
=> s ferrocen? (s) (prote##### or peptidase or hydrol#### or cleav### or
digest####)
```

```
5 FILE AGRICOLA
71 FILE ANABSTR
3 FILE ANTE
45 FILE BIOENG
56 FILE BIOSIS
65 FILE BIOTECHABS
65 FILE BIOTECHDS
30 FILE BIOTECHNO
```

13 FILES SEARCHED...

```
14 FILE CABA
918 FILE CAPLUS
17 FILE CEABA-VTB
1 FILE CONFSCI
4 FILE DDFB
14 FILE DDFU
16 FILE DGENE
```

23 FILES SEARCHED...

```
57 FILE DISSABS
4 FILE DRUGB
20 FILE DRUGU
3 FILE EMBAL
84 FILE EMBASE
81 FILE ESBIOWASE
3 FILE FROSTI
5 FILE FSTA
```

35 FILES SEARCHED...

```
1 FILE HEALSAFE
```

116 FILE IFIPAT
 43 FILE LIFESCI
 85 FILE MEDLINE
 19 FILE NTIS
 227 FILE PASCAL
 47 FILES SEARCHED...
 2 FILE PHIN
 11 FILE PROMT
 288 FILE SCISEARCH
 7 FILE SYNTHLINE
 51 FILE TOXCENTER
 59 FILES SEARCHED...
 1020 FILE USPATFULL
 32 FILE USPATOLD
 230 FILE USPAT2
 168 FILE WPIDS
 2 FILE WPIFV
 168 FILE WPINDEX

40 FILES HAVE ONE OR MORE ANSWERS, 68 FILES SEARCHED IN STNINDEX

L1 QUE FERROCEN? (S) (PROTE##### OR PEPTIDASE OR HYDROL#### OR CLEAV### OR DIGEST####)

=> s L1 (s) (mark## or label### or tag###)

1 FILE AGRICOLA
 15 FILE ANABSTR
 12 FILE BIOENG
 8 FILE BIOSIS
 17 FILE BIOTECHABS
 17 FILE BIOTECHDS
 6 FILE BIOTECHNO
 13 FILES SEARCHED...
 2 FILE CABA
 47 FILE CAPLUS
 3 FILE CEABA-VTB
 1 FILE DDFU
 13 FILE DGENE
 23 FILES SEARCHED...
 3 FILE DISSABS
 3 FILE DRUGU
 1 FILE EMBAL
 6 FILE EMBASE
 10 FILE ESBIODBASE
 1 FILE FSTA
 35 FILES SEARCHED...
 32 FILE IFIPAT
 14 FILE LIFESCI
 15 FILE MEDLINE
 24 FILE PASCAL
 47 FILES SEARCHED...
 17 FILE SCISEARCH
 8 FILE TOXCENTER
 88 FILE USPATFULL
 60 FILES SEARCHED...
 7 FILE USPAT2
 14 FILE WPIDS
 14 FILE WPINDEX

28 FILES HAVE ONE OR MORE ANSWERS, 68 FILES SEARCHED IN STNINDEX

L2 QUE L1 (S) (MARK## OR LABEL### OR TAG###)

=> s L2 (s) substrat#

```
      3  FILE ANABSTR
      1  FILE BIOENG
      5  FILE BIOTECHABS
      5  FILE BIOTECHDS
      2  FILE BIOTECHNO
13 FILES SEARCHED...
      2  FILE CAPLUS
      2  FILE DGENE
      1  FILE DISSABS
27 FILES SEARCHED...
      2  FILE ESBIOBASE
      1  FILE FSTA
     13  FILE IFIPAT
37 FILES SEARCHED...
      2  FILE LIFESCI
      2  FILE PASCAL
47 FILES SEARCHED...
     19  FILE USPATFULL
      1  FILE USPAT2
62 FILES SEARCHED...
      5  FILE WPIDS
      5  FILE WPINDEX
```

17 FILES HAVE ONE OR MORE ANSWERS, 68 FILES SEARCHED IN STNINDEX

L3 QUE L2 (S) SUBSTRAT#

=> d rank

```
F1      19  USPATFULL
F2      13  IFIPAT
F3       5  BIOTECHABS
F4       5  BIOTECHDS
F5       5  WPIDS
F6       5  WPINDEX
F7       3  ANABSTR
F8       2  BIOTECHNO
F9       2  CAPLUS
F10      2  DGENE
F11      2  ESBIOBASE
F12      2  LIFESCI
F13      2  PASCAL
F14      1  BIOENG
F15      1  DISSABS
F16      1  FSTA
F17      1  USPAT2
```

=> fil f3-f9, f11, f12

COST IN U.S. DOLLARS

SINCE FILE	TOTAL
ENTRY	SESSION
10.88	11.10

FULL ESTIMATED COST

FILE 'BIOTECHABS' ACCESS NOT AUTHORIZED

FILE 'BIOTECHDS' ENTERED AT 09:25:07 ON 09 MAR 2009
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FILE 'WPIDS' ENTERED AT 09:25:07 ON 09 MAR 2009

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FILE 'WPINDEX' ACCESS NOT AUTHORIZED

FILE 'ANABSTR' ENTERED AT 09:25:07 ON 09 MAR 2009
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FILE 'BIOTECHNO' ENTERED AT 09:25:07 ON 09 MAR 2009
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FILE 'CAPLUS' ENTERED AT 09:25:07 ON 09 MAR 2009
USE IS SUBJECT TO THE TERMS OF YOUR STN CUSTOMER AGREEMENT.
PLEASE SEE "HELP USAGETERMS" FOR DETAILS.
COPYRIGHT (C) 2009 AMERICAN CHEMICAL SOCIETY (ACS)

FILE 'ESBIOBASE' ENTERED AT 09:25:07 ON 09 MAR 2009
COPYRIGHT (C) 2009 Elsevier Science B.V., Amsterdam. All rights reserved.

FILE 'LIFESCI' ENTERED AT 09:25:07 ON 09 MAR 2009
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=> s L3
4 FILES SEARCHED...
L4 21 L3

=> dup rem L4
PROCESSING COMPLETED FOR L4
L5 18 DUP REM L4 (3 DUPLICATES REMOVED)

=> s L5 and py<2004
L6 8 L5 AND PY<2004

=> d L6 ibib abs 1-8

L6 ANSWER 1 OF 8 BIOTECHDS COPYRIGHT 2009 THOMSON REUTERS on STN
ACCESSION NUMBER: 2004-09731 BIOTECHDS <<LOGINID::20090309>>
TITLE: Detecting biomolecules e.g. DNA, for disease diagnosis,
comprises applying biochemical probe to substrate with
biodetection site including electronic resonator or reacting
biomolecule with magnetic tag;
DNA biosensor, electrical resonator and DNA probe or RNA
probe for nucleic acid detection
AUTHOR: BEUHLER A; SHMAGIN I; LACH L
PATENT ASSIGNEE: BEUHLER A; SHMAGIN I; LACH L
PATENT INFO: US 2003027148 6 Feb 2003
APPLICATION INFO: US 2001-916108 26 Jul 2001
PRIORITY INFO: US 2001-916108 26 Jul 2001; US 2001-916108 26 Jul 2001
DOCUMENT TYPE: Patent
LANGUAGE: English
OTHER SOURCE: WPI: 2004-118833 [12]
AN 2004-09731 BIOTECHDS <<LOGINID::20090309>>
AB DERWENT ABSTRACT:
NOVELTY - Detecting a biomolecule comprises applying biochemical probe to
substrate with biodetection site including electronic resonator
or reacting biomolecule with magnetic tag and contacting the
tagged biomolecule to biodetection site on resonator and
detecting the number of biomolecules.
DETAILED DESCRIPTION - INDEPENDENT CLAIMS are also included for: (1)
Detecting a biomolecule comprising: (a) providing a substrate
with biodetection site(s) including an electrical resonator; applying

biochemical probe(s) to the biodetection site; (b) interacting the biomolecule with the biochemical probe at the biodetection site; and (c) detecting the biomolecule at the biodetection site; (2) Detecting a biomolecule comprises reacting a biomolecule with a magnetic tag, contacting the tagged biomolecule to a biodetection site on a resonator, coupling the biomolecule to probes in solution and detecting the number of biomolecules; (3) A biosensor for assaying biomolecules, comprising: (a) a substrate having at least one biodetection site; and (b) an electrical resonator proximate the biodetection site to allow measurement of magnetic properties at the biodetection site; and (4) A kit for assaying target biomolecules comprising: (a) a sample plate with biodetection sites; (b) a holding gel adapted to coat the biodetection sites; and (c) an electrical resonator operatively attached to the sample plate to allow measurement of magnetic properties of the biodetection sites.

BIOTECHNOLOGY - Preferred Method: The method further comprises applying a magnetic tag to the biomolecule and detecting magnetic properties of the biodetection site with the resonator. The value of the magnetic properties of the biodetection site corresponds to a number of biomolecules at the biodetection site. The method further comprises applying a holding substance to the biodetection site. The substrate comprises an organic substrate with embedded resonance frequency structures. The biochemical probe is an oligonucleotide. The magnetic tag is ferrocene. The magnetic properties of the biodetection site are measured before applying the biomolecule. The method further comprises determining an alteration in magnetic properties of the biodetection site. The value of the alteration in magnetic properties with a number of biomolecules is correlated and the number of biomolecules at the biodetection site is determined based upon this value of alteration. **Preferred Biosensor:** The biodetection site includes a holding substance. The biodetection site is adapted to receive biomolecule(s) and further comprises magnetic tag(s) operatively adapted to be applied to the biomolecule. The electrical resonator measures a value of magnetic properties, the value corresponding to a number of biomolecules at the biodetection site. The electrical resonator has a spiral geometry. It further comprises a measuring component for measuring the magnetic properties in communication with the biodetection site and a quantitating element for correlating an amount of biomolecules at the biodetection site to the magnetic properties of the biodetection site. **Preferred Kit:** The kit comprises a solution of biochemical probe(s) adapted to be applied to the biodetection sites and a solution of magnetic tag(s) adapted to be applied to the target biomolecules.

USE - The methods and kits are useful for detecting biomolecules such as DNA and RNA.

EXAMPLE - A substrate containing multiple surface resonator test sites with fully defined geometry and specifications was fabricated by using standard printed circuit board print and etch technology. A polymer hydrogel coating containing biomolecular probe linker groups was draw coated onto the surface of 10 of the test sites to a thickness of 5 microns. The polymer coating was exposed to UV radiation of 1000 mJ/cm² at 365 nm to crosslink the polymer and render it insoluble to water solutions. 40 microL of a 100 microgram/ml streptavidin protein was then washed over the surface of 5 of the resonators. The resonators were irradiated again with UV light at 365 nm (100 mJ/cm²) to photoactively couple the streptavidin to the probe linker groups in the polymer coating. The resonators were washed to remove excess material. The resulting protein concentration on the surface was approximately 0.67 nmoles per test site. The resonance frequency of the 5 test sites with the bound streptavidin were compared to 5 test sites with no streptavidin. The test sites containing streptavidin showed

a 14 MHz frequency shift compared to the test sites with no streptavidin. This indicates that 0.67 nmoles per test site of a large protein such as streptavidin can be detected. Each test site is 1.94 cm² so this is a sensitivity of 0.35 nmoles/cm². In this case the frequency shift is due to the change in the capacitive properties of the test site due to the presence of streptavidin since the streptavidin does not contain a magnetic tag.(12 pages)

L6 ANSWER 2 OF 8 BIOTECHDS COPYRIGHT 2009 THOMSON REUTERS on STN

ACCESSION NUMBER: 2003-12443 BIOTECHDS <<LOGINID::20090309>>

TITLE: Method for detecting ligand-ligate interaction, useful e.g. in immunological or hybridization assays, based on displacement of labeled signal ligand; ligand-ligate interaction and biochip useful for hybridization assay

AUTHOR: HARTWICH G; FRISCHMANN P; HAKER U; WIEDER H

PATENT ASSIGNEE: FRIZ BIOCHEM GMBH

PATENT INFO: WO 2003019194 6 Mar 2003

APPLICATION INFO: WO 2002-DE1269 6 Apr 2002

PRIORITY INFO: DE 2001-1041691 25 Aug 2001; DE 2001-1041691 25 Aug 2001

DOCUMENT TYPE: Patent

LANGUAGE: German

OTHER SOURCE: WPI: 2003-290094 [28]

AN 2003-12443 BIOTECHDS <<LOGINID::20090309>>

AB DERWENT ABSTRACT:

NOVELTY - Method for detecting ligate-ligand interactions by contacting a surface, modified by attachment of at least one ligate (L1), with a known amount of signal ligand (SL) and a sample containing a ligand (L2). SL are detected and the result compared with a reference value.

DETAILED DESCRIPTION - An INDEPENDENT CLAIM is also included for a kit for the new method.

USE - The method is used to detect e.g. antibody/antigen; nucleic acid/nucleic acid or enzyme/substrate, or many other sorts of interactions, particularly with low-density protein/DNA chips for point-of-care systems.

ADVANTAGE - The method is a simple, inexpensive and reliable displacement assay that does not require modification of the target by labeling.

EXAMPLE - An oligonucleotide was modified with the residue HO-(CH₂)-S-(CH₂)₂- and immobilized on gold. A complementary nucleic acid tetramer, labeled twice with ferrocene, (10 microM) was applied and chronocoulometric measurements (1) made. The complementary target (5 microM) was added and the measurements (2) repeated. The plots of charge against square root of time are presented and are clearly different between (1) and (2), with the integrated difference between the two curves being 0.7 microC.(57 pages)

L6 ANSWER 3 OF 8 BIOTECHDS COPYRIGHT 2009 THOMSON REUTERS on STN

ACCESSION NUMBER: 2002-10819 BIOTECHDS <<LOGINID::20090309>>

TITLE: Interaction of species (e.g. biological ligand) immobilized on colloidal particles, with species (e.g. molecule capable of reacting with ligand) immobilized on non-colloidal structure e.g. magnetic beads, useful in e.g. drug screening; biological ligand interaction, colloid particle immobilization and magnetic bead, for drug screening

AUTHOR: BAMDAD C C; BAMDAD R S

PATENT ASSIGNEE: MINERVA BIOTECHNOLOGIES CORP

PATENT INFO: WO 2002001228 3 Jan 2002

APPLICATION INFO: WO 2000-US20168 23 Jun 2000

PRIORITY INFO: US 2000-602778 23 Jun 2000

DOCUMENT TYPE: Patent

LANGUAGE: English
OTHER SOURCE: WPI: 2002-205935 [26]
AN 2002-10819 BIOTECHDS <<LOGINID::20090309>>
AB DERWENT ABSTRACT:

NOVELTY - (1) A method comprises allowing a colloid particle the ability to become immobilized with respect to a non-colloidal structure (II); and determining immobilization of the colloid particle relative to (II).

DETAILED DESCRIPTION - INDEPENDENT CLAIMS are also included for the following: (2) signaling a single binding of a first biological or chemical agent to a second biological or chemical agent with a plurality of signaling entities; (3) determining protein/ligand interaction in the absence of SPR without labeling either the protein or the ligand; (4) a method, comprising: (a) providing: (i) a solution comprising colloids, the colloids comprising a ligand capable of interacting with a cell surface molecule and (ii) a composition comprising an electrode comprising growing cells, the cells comprising at least one cell surface molecule capable of interacting with the ligand; (b) adding at least a portion of the colloids to the composition; (5) a method comprising: (a1) providing: (i) a solution comprising colloids, the colloids comprising a ligand capable of interacting with a cell surface molecule, (ii) a candidate drug, and (iii) a composition comprising an electrode comprising growing cells, the cells comprising at least one cell surface molecule capable of interacting with the ligand; and (b1) mixing at least a portion of the colloids with the drug and the composition; (6) recruiting an electronic signaling entity to an electrode using a magnetic material; (7) an article defining a surface, and a ligand suspected of interacting with a protein and an electroactive entity each immobilized relative to the surface; (8) an article comprising: a first biological or chemical agent, capable of biological or chemical binding to a second agent, immobilized relative to a plurality of signaling entities; (9) an article defining a surface, and a self-assembled monolayer formed on the surface of the article; (10) a composition comprising a first molecule and one or more signaling entities attached to a solid support, where the first molecule is a ligand capable of interacting with a cell-surface receptor or protein; (11) a composition, comprising a first molecule, a second molecule and a third molecule attached to a solid support, where the first molecule comprises a ligand capable of interacting with a cell-surface receptor or protein, where the second molecule forms a monolayer on the solid support, and where the third molecule is electroactive; (12) an article comprising a metal support constructed and arranged to support the growth of cells on its surface, the metal support comprising a monolayer of at least one type of molecule, the monolayer configured such that the metal support can be used as an electrode; (13) a composition comprising: a colloid particle; a signaling entity immobilized relative to the colloid particle; and a protein immobilized relative to the colloid particle; (14) a species comprising: a polymer or dendrimer carrying a plurality of signaling entities adapted for linkage to a biological or chemical agent (I); (15) an article comprising a colloid particle immobilized relative to a glutathione derivative and at least one signaling entity; (16) an article comprising a colloid particle carrying on its surface, a self-assembled monolayer comprising a glutathione derivative.

USE - Methods, compositions, species and articles for detecting or monitoring interactions between chemical and biochemical species, including drug screening assays, are provided. For detecting interactions between ligands and target receptors on the surface of live intact cells to enable screening of candidate compounds which disrupt these interactions. For screening compound libraries for drugs that inhibit the activity of cell surface receptors. For examining e.g. protein/protein, protein/peptide, antibody/antigen,

antibody/hapten, enzyme/substrate, enzyme/inhibitor, enzyme/cofactor, binding protein/substrate, carrier protein/substrate, lectin/carbohydrate, receptor/hormone, receptor/effector, complementary strands of nucleic acid, protein/nucleic acid, repressor/inducer, ligand/cell surface receptor and virus/ligand interactions. For identifying cell-derived molecules (e.g. receptors or proteins), that are expressed differentially in healthy versus diseased tissue or cells, i.e. diagnostic assays for determination of diseased states. For visually investigating patterns of cell surface receptor expression on individual cell surfaces and/or on cells embedded in a tissue specimen. Biospecific colloids (gold colloids) may be used to facilitate in vivo imaging, e.g. detection of tumor using X-ray or X-ray computer tomography.

ADVANTAGE - A series of components and techniques for drug screening are provided. The approach provides: (i) modular system for the attachment of natural ligands to universal signaling elements; (ii) enhanced sensitivity of detection through the attachment of a plurality of signaling elements to each ligand; (iii) a simpler format (without the need for washing steps, enzymatic cleavage and toxic substrates); (iv) a convenient electronic output; and (v) the capability of multiplexing. Further advantages over existing methods such as ELISA, fluorescent labeling and SPR include: in the above systems, there is no need for protein labeling; the protein is attached to a labeled component. Gold colloids can be pre-labeled with both: (a3) a signaling moiety; and (b3) a functional group for protein attachment. Self assembled monolayers that present both NTA/Ni²⁺, to capture histidine-tagged proteins, and a ferrocene derivative, for electronic or electrochemical signaling, can be formed on the colloids. SAMs that incorporate carboxylic acid groups, for the chemical coupling (standard EDC/NHS chemistry) of unmodified proteins, can also be used. Virtually any biological species can be co-immobilized on colloids with a signaling entity. The technology enables cost-effective multiplexing as it can readily be multiplexed on microelectrode arrays. (NTA = nitrilo triacetic acid; SAM = self-assembled monolayer).

EXAMPLE - A target protein, Glutathione-S-Transferase (GST) was histidine-tagged and immobilized on SAM-coated colloids that presented NTA-Ni (histamine tags bind NTA-Ni). 30 microl of colloids presenting 40 microM NTA-Ni on the surface were added to 65 microl of 21.5 microM GST, to give a final concentration of 14 microM GST in solution. Glutathione, a small molecule that binds GST, is commercially available bound to agarose beads through Sigma-Aldrich. Glutathione-coated beads were incubated with the solution of GST-bound colloids. Within minutes, the GST bound to the glutathione beads, bringing the colored colloids out of solution, and decorating the beads red (Figure 20). Beads displaying a small molecule that does not bind to GST remained colorless when incubated with the GST-bound colloids (Figure 21). A second negative control, in which glutathione-coated beads were incubated with 30 microl NTA-Ni colloids in the absence of GST showed that NTA-Ni-colloids do not bind nonspecifically to the bead surfaces or to the glutathione. (83 pages)

L6 ANSWER 4 OF 8 ANABSTR COPYRIGHT 2009 RSC on STN
 AB Biotin (I) is determined by addition to a mixture containing trypsin-labelled I that is inhibited by avidin; I reacts with the avidin to release the trypsin - I, which initiates a two-step enzyme cascade in which chymotrypsinogen is activated to chymotrypsin. The chymotrypsin hydrolyses dimethylferrocenoylated Tyr-Gly-Gly immobilized on AH-Sepharose 4B to release ferrocenoyltyrosine, which is oxidized at a vitreous-carbon electrode in the presence of glucose oxidase

and D-glucose; 20mM-K₂HPO₄ (pH 6.9) is used as supporting electrolyte. The catalytic current produced (measured by using a Pt-gauze counter-electrode and a SCE) is related rectilinearly to the I concentration (0.4 to 5µM) if the enzyme reactions are not substrate-limited. The magnitude of response is time-dependent.

L6 ANSWER 5 OF 8 BIOTECHNO COPYRIGHT 2009 Elsevier Science B.V. on STN
ACCESSION NUMBER: 2000:32002327 BIOTECHNO <<LOGINID::20090309>>
TITLE: Immunoassay of the MRSA-related toxic protein, leukocidin, with scanning electrochemical microscopy
AUTHOR: Kasai S.; Yokota A.; Zhou H.; Nishizawa M.; Niwa K.; Onouchi T.; Matsue T.
CORPORATE SOURCE: T. Matsue, Dept. of Biomolecular Engineering, Graduate School of Engineering, Tohoku University, Sendai 980-8579, Japan.
SOURCE: Analytical Chemistry, (01 DEC 2000), 72/23 (5761-5765), 36 reference(s)
CODEN: ANCHAM ISSN: 0003-2700
DOCUMENT TYPE: Journal; Article
COUNTRY: United States
LANGUAGE: English
SUMMARY LANGUAGE: English

AN 2000:32002327 BIOTECHNO <<LOGINID::20090309>>
AB Scanning electrochemical microscopy (SECM) was applied to the immunoassay of leukocidin, which is a toxic protein produced by methicillin-resistant *Staphylococcus aureus* (MRSA), with the intention of developing an early diagnostic for MRSA infection. An antibody - chip for leukocidin was prepared by self-assembling of anti-leukocidin on a protein A-coated glass substrate. A sample solution containing leukocidin was spotted onto the antibody-chip, followed by labeling with horseradish peroxidase (HRP) via a sandwich method. The reduction current of the oxidized form of ferrocenylmethanol generated by the HRP reaction was monitored to view SECM images of the spot of captured leukocidin. The amplitude of reduction current depended on the concentrations of sample solutions used for making spots. This SECM-based immunoassay detects as low as 5.25 pg mL⁻¹.
leukocidin.

L6 ANSWER 6 OF 8 BIOTECHNO COPYRIGHT 2009 Elsevier Science B.V. on STN
ACCESSION NUMBER: 1996:27037958 BIOTECHNO <<LOGINID::20090309>>
TITLE: Enzyme immunoassay with amperometric flow-injection analysis using horseradish peroxidase as a label. Application to the determination of polychlorinated biphenyls
AUTHOR: Del Carlo M.; Mascini M.
CORPORATE SOURCE: M. Mascini, Dipartimento Sanita Pubblica, Epidemiol/Chimica Analit Ambientale, Universita degli Studi di Firenze, Via G. Capponi 9, 50121 Firenze, Italy.
E-mail: mascini@cesit1.unifi.it.
SOURCE: Analytica Chimica Acta, (1996), 336/1-3 (167-174), 25 reference(s)
CODEN: ACACAM ISSN: 0003-2670
PUBLISHER ITEM IDENT.: S0003267096003777
DOCUMENT TYPE: Journal; Article
COUNTRY: Netherlands
LANGUAGE: English
SUMMARY LANGUAGE: English
AN 1996:27037958 BIOTECHNO <<LOGINID::20090309>>
AB An amperometric detection system for horseradish peroxidase (HRP) activity was optimized using flow injection analysis (FIA) with glassy

carbon as a working electrode. Ferroceneacetic acid was investigated as a co-substrate for the electrochemical detection of HRP. The calculated detection limit for HRP was 2.6×10^{-10} M with incubation of 30 min. The substrate was used in an electrochemical enzyme immunoassay for polychlorinated biphenyls (PCB). We used a competitive assay, where PCB-protein conjugate (gelatin) was immobilized to the solid phase (microtitre assay plate) and the competition was carried out with PCB standards using a limiting amount of anti-PCB IgG. The extent of the competition was evaluated using a secondary, HRP labelled, IgG; the amount of the enzyme label was detected after 30 min of incubation with the substrate. The PCB range was 0.1–50 $\mu\text{g ml}^{-1}$. The overall assay time was 2 h and 30 min. The within assay precision, over 6 measurements, was lower than 10% for the entire range.

L6 ANSWER 7 OF 8 Elsevier Biobase COPYRIGHT 2009 Elsevier Science B.V. on STN

ACCESSION NUMBER: 1998022420 ESBIODASE <<LOGINID::20090309>>
 TITLE: Covalent attachment of an electroactive sulfhydryl reagent in the active site of cytochrome P450(cam) as revealed by the crystal structure of the modified protein
 AUTHOR(S): Di Gleria, Katalin; Nickerson, Darren P.; Hill, H. Allen O.; Wong, Luet-Lok; Fulop, Vilmos
 CORPORATE SOURCE: Di Gleria, Katalin; Nickerson, Darren P.; Hill, H. Allen O.; Wong, Luet-Lok (Department of Chemistry, Inorganic Chemistry Laboratory, University of Oxford, South Parks Road, Oxford OX1 3QR (GB)); Fulop, Vilmos (Laboratory of Molecular Biophysics, Oxford Centre for Molecular Sciences, University of Oxford, South Parks Road, Oxford OX1 3QU (GB))
 SOURCE: Journal of the American Chemical Society (14 Jan 1998) Volume 120, Number 1, pp. 46–52, 31 refs.
 CODEN: JACSAT ISSN: 0002-7863
 DOI: 10.1021/ja972473k
 COUNTRY OF PUBLICATION: United States of America
 DOCUMENT TYPE: Journal; Article
 LANGUAGE: English
 SUMMARY LANGUAGE: English
 ENTRY DATE: Entered STN: 31 Jan 2009
 Last updated on STN: 31 Jan 2009

AN 1998022420 ESBIODASE <<LOGINID::20090309>>

AB A novel electroactive sulfhydryl-specific reagent, N-(2-ferrocenylethyl)maleimide (Fc-Mi), was used to attach a redox-active reporter group to cytochrome P450(cam) from *Pseudomonas putida*. The crystal structure of the modified enzyme was determined at 2.2 Å resolution ($R(\text{cryst}) = 0.18$) and compared to the structure of the wild-type enzyme complexed with its natural substrate. The results showed that two molecules of the electroactive modifier were attached to the protein. One of the ferrocenes was linked to Cys85 via the maleimide moiety and occupied the camphor-binding site in the substrate pocket. The other ferrocene was linked to Cys136 on the surface of the protein. Significant conformational changes were observed on the distal side of the heme when camphor was replaced by ferrocene. The shift in the Soret band from 392 to 417 nm upon modification arose from the binding of a water molecule to the heme iron immediately below the ferrocene in the active site of the modified enzyme. The electrochemistry of the labeled enzyme showed clear signals originating both from the heme and from the covalently linked ferrocenes. The direct current cyclic

voltammogram revealed a striking positive shift in the heme redox potential of the ferrocene-containing P450(cam) from -380 mV for the camphor-bound wild-type protein to -280 mV for the modified protein.

L6 ANSWER 8 OF 8 LIFESCI COPYRIGHT 2009 CSA on STN
ACCESSION NUMBER: 2000:72742 LIFESCI <<LOGINID::20090309>>
TITLE: Covalent Attachment of an Electroactive Sulfhydryl Reagent in the Active Site of Cytochrome P450 sub(cam) as Revealed by the Crystal Structure of the Modified Protein
AUTHOR: Di Gleria, K.; Nickerson, D.P.; Hill, H.A.O.; Wong, L.-L.; Fueloep, V.
CORPORATE SOURCE: Department of Chemistry, Inorganic Chemistry Laboratory, University of Oxford, South Parks Road, Oxford OX1 3QR, UK
SOURCE: Journal of the American Chemical Society [J. Am. Chem. Soc.], (19980100) vol. 120, no. 1, pp. 46-52. ISSN: 0002-7863.
DOCUMENT TYPE: Journal
FILE SEGMENT: J
LANGUAGE: English
SUMMARY LANGUAGE: English
AB A novel electroactive sulfhydryl-specific reagent, N-(2-ferrocenylethyl)maleimide (Fc-Mi), was used to attach a redox-active reporter group to cytochrome P450 sub(cam) from *Pseudomonas putida*. The crystal structure of the modified enzyme was determined at 2.2 Angstrom resolution ($R_{\text{sub}(\text{cryst})} = 0.18$) and compared to the structure of the wild-type enzyme complexed with its natural substrate. The results showed that two molecules of the electroactive modifier were attached to the protein. One of the ferrocenes was linked to Cys85 via the maleimide moiety and occupied the camphor-binding site in the substrate pocket. The other ferrocene was linked to Cys136 on the surface of the protein. Significant conformational changes were observed on the distal side of the heme when camphor was replaced by ferrocene. The shift in the Soret band from 392 to 417 nm upon modification arose from the binding of a water molecule to the heme iron immediately below the ferrocene in the active site of the modified enzyme. The electrochemistry of the labeled enzyme showed clear signals originating both from the heme and from the covalently linked ferrocenes. The direct current cyclic voltammogram revealed a striking positive shift in the heme redox potential of the ferrocene-containing P450 sub(cam) from -380 mV for the camphor-bound wild-type protein to -280 mV for the modified protein.

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